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Developments in Irradiation of Fresh Fruits and Vegetables

New studies show the potential of irradiation for control of plant and human pathogens associated with fresh fruits and vegetables

DONALD W. THAYER AND KATHLEEN T. RAJKOWSKI

Author Thayer, a Professional Member of IFT, is Research Leader and author Rajkowski is Research Chemist, U.S. Dept. of Agriculture, Agricultural Research Service, Eastern Regional Research Center, 600 East Mermaid Lane, Wyndmoor, PA 19038. Send reprint requests to author Thayer.

The application of ionizing irradiation to control spoilage microorganisms increases shelf life of irradiated strawberries, lettuce, sweet onions, and carrots.

Several studies have been published on the irradiation of seed to control plant pathogens. To date, relatively little effort has been applied to the control of foodborne pathogens on fresh foods. However, ionizing irradiation has recently been used to eliminate *Escherichia coli* O157:H7 from apple juice, *Toxoplasma gondii* and/or *Cyclospora cayetanensis* from raspberries, and *E. coli* O157:H7 and salmonellae from seed and sprouts. Studies are underway in several countries to find suitable methods to control human foodborne pathogens associated with fresh fruits, fruit juices, fresh cut vegetables or salads, sprouts, and seed. Combination treatments with ionizing radiation and other processes such as chlorination seem especially promising.

Foodborne Illness Outbreaks

Ingestion of raw vegetables, fruits, and fruit juices have been linked to outbreaks of foodborne illness. Contaminated artichoke, beet leaves, cabbage, carrots, cauliflower, celery, cucumber, egg plant, endive, fennel, onion, mustard cress, lettuce, mushrooms, parsley, pepper, potatoes, prepacked salads, salad vegetables, spinach, sprouts, and tomatoes have all been vehicles for transmission of pathogens (Beuchat, 1996). The pathogens identified with these outbreaks include *Aeromonas, Bacillus cereus, Campylobacter jejuni, enterotoxigenic E. coli* and *E. coli* O157:H7, *Salmonella, Listeria monocytogenes, Shigella, Staphylococcus, Staphylococcus aureus, Vibrio cholerae, Yersinia enterocolitica, hepatitis A*, and Norwalk/Norwalk-like virus (Beuchat, 1996). Apple cider, cantaloupe, orange juice, raspberries, strawberries, and watermelon have been associated withoutbreaks of disease caused by *Cyclospora cayetanensis, Cryptosporidium parvum, E. coli* O157:H7, *hepatitis A*, Norwalk/Norwalk-like virus, and *Salmonella spp.* (Tauxe et al., 1997).

Consumption of raw vegetable sprouts has been linked to several outbreaks of foodborne illness. In 1995, there was a major outbreak of illness caused by alfalfa sprouts, possibly triggered by *Salmonella newport* in Denmark, Canada, and the United States associated with the same lot of alfalfa seeds (Aabo and Baggesen, 1997; Beneden et al., 1999). The consumption of radish sprouts contaminated with *E. coli* O157:H7 was linked to nearly 6,000 cases of illness in Sakai City, Japan, in 1996 (WHO, 1997). During 1998, California restricted the sale of raw sprouts after three sprout-related outbreaks due to *Salmonella* and *E. coli* O157:H7. In each of these cases, seeds were the suspect carrier of the pathogen. In 1999, outbreaks of *Salmonella mbandaka* associated with the ingestion of alfalfa sprouts occurred in Idaho, Oregon, and Washington and were linked to contaminated seed from southern California (Anonymous. 1999). On July 9, 1999, the Food and Drug Administration issued the following warning: "Because of reports of increasing numbers of illnesses associated with consumption of raw sprouts, the Food and Drug Administration is advising all persons to be aware of the risks associated with eating raw sprouts (e.g., alfalfa, clover, radish). Outbreaks have included persons of both genders and all age categories. Those persons who wish to reduce the risk of foodborne illness from sprouts are advised not to eat raw sprouts."

In contrast to the extensive studies on irradiation to control pathogens on meat and poultry products, there are very few studies of the value of ionizing irradiation for the elimination of foodborne pathogens on or in fruit juice; fruits; vegetables, such as lettuce and sprouts; and seed used to grow sprouts. These foods are often eaten raw without benefit of any pathogen killing step. Most studies of the irradiation of fruits and vegetables have not considered the possibility of controlling human pathogens, and only a very few studies considered the effects of controlling temperature or of using modified-atmosphere packaging (MAP) on the effectiveness of the process or in quality preservation.

Leafy vegetables, such as lettuce, have been considered unsuitable for irradiation because of phytotoxic damage that was encountered during early studies with high-dose irradiation intended to produce a sterile product. MAP is very frequently used to extend the shelf life of sprouts and precut salad mixtures. Unfortunately, both of these products have been linked to outbreaks of foodborne disease in the United States caused by *E. coli* O157:H7, *Salmonella*, and *L. monocytogenes*. Treatment with disinfectants including hypochlorite have not completely eliminated these pathogens from seeds, nor have they been very effective in the decontamination of sprouts (Beuchat, 1997). The sprouts may be contaminated internally and are relatively fragile and not likely to respond well to chemical decontamination.

During 1991, there was an outbreak of diarrhea and hemolytic uremic syndrome from *E. coli* O157:H7 in fresh apple juice (Besser et al., 1993). During 1996, unpasteurized apple cider was associated with three outbreaks of *E. coli* O157:H7 illness involving 66 persons and 1 death in Connecticut and New York that were traced to unpasteurized apple cider (MMWR, 1997).

Salmonella have been associated with the ingestion of unpasteurized orange juice (Cook et al., 1998).

Basic studies are needed for fresh fruits and vegetables, including the optimum time of harvest, the specific varieties which best withstand treatment with radiation, the doses required to inactivate the common pathogens, the factors that may alter the percentage killed, and the interactions of radiation injury of the pathogen with other treatments. The use of combination treatments is expected to be more effective both in eliminating pathogens and in retaining quality attributes of the product.

The irradiation of grain, fruits, and vegetables is approved by FDA to a maximum dose of 1 kGy for disinfestation. Fruits and vegetables may also be irradiated to inhibit sprouting and to alter the rates of maturation to obtain greater shelf life. The current regulations (21 CFR Part 179.26) do not include the use of irradiation to control foodborne pathogens on fruits and vegetables.

Irradiation Studies

The purposes of most previous research studies on the irradiation of fruits and vegetables were to alter rates of ripening, control postharvest pathogens, and disinfest. Though these studies did not include human pathogens, they provided data on the radiation doses that these products will tolerate and also on the doses required to inactivate plant pathogens. These areas have been the subject of several symposia and many reviews. The data up to 1988 were reviewed by Thomas (1983, 1984ab, 1985, 1986, 1988). The International Atomic Energy Agency of the Food and Agriculture Organization has sponsored several symposia and workshops on insect disinfestation (ICGFI/IAEA, 1991). Willemot et al. (1996) reviewed the preservation of fruits with ionizing radiation.

• **Fruits and Juices.** The control of postharvest fungal spoilage of fruits by *Botrytis cinerea* has been the subject of many studies. Maxie (1968) observed that *B. cinerea* could be better controlled with 0.1 kGy if there was a prior heat sensitization than by 200 Mrad with heat sensitization. Barkai et al. (1971) observed that a dose of 0.2 kGy was more effective than 0.1 kGy in reducing decay in strawberries during storage. El Sayed (1978) observed that 0.1 and 0.2 kGy gamma-ray doses of soft-type date fruits suppressed fungal and bacterial spoilage. Tiryaki et al. (1994) discovered that doses of 1, 2, 3, and 3.5 kGy did not completely control postharvest decay of apple, quince, onion, and peach, but did delay the growth of *Penicillium expansum*, *Monilinia fructigena*, *Botrytis aclada*, and *Rhizopus stolonifer*. Yu et al. (1995) found that irradiating strawberries with doses of 1 and 2 kGy of electrons suppressed fungi on stored berries and increased storage time.

Studies of irradiated fruits included the juices and provided information on the tolerance of these products to ionizing radiation. Proctor and O'Meara (1951) studied the effects of ionizing radiation on the ascorbic acid content of orange juice and found that when the juice was frozen the loss was negligible. Bregvadze (1963) found that a dose of 0.5 kGy completely sterilized apple juice, which kept its characteristic odor and taste. Fetter et al. (1969) found that doses up to a maximum of 5 kGy had no effect on the flavor of commercial orange, guava, tomato, redcurrant, blackcurrant, apricot, peach, pear, and grape juice. Kaupert et al. (1981) found that *Saccharomyces rouxii* was eliminated from apple and pear juice concentrates by a dose of 0.4 kGy. Kiss and Farkas (1970) observed marked increases in storage life of Jonathan apple juice concentrates by irradiation. A dose of 13 kGy did not affect taste or aroma and ensured a storage life of at least 10 mo at room temperature. Partsch et al. (1970) inoculated apple, grape, and orange juices with *Byssochlamys fulva* and investigated the effects of irradiation at 50 and 72°C.

At 72°C, the lethal doses were 0.16, 0.18, and 0.18 kGy in apple, grape, and orange juices, respectively. Buchanan et al. (1998) found that the D-value for *E. coli* O157:H7 in apple juice at 2°C was dependent on the level of suspended solids and ranged from 0.26 to 0.35 kGy. The authors concluded that a dose of 1.8 kGy should be sufficient to achieve a 5D inactivation of *E. coli* O157:H7. Buchanan et al. (1999) discovered that acid adaption of *E. coli* O157:H7 increased its resistance to radiation.

• **Cells and Seeds.** Research on the effects of ionizing radiation on plant cells and seeds started early in the century, when Guilleminot (1908) exposed pumpkin seed to x-rays and observed retardation of growth and decreased viability of the plants. Osborne et al. (1963) found that when pre-irradiation humidity level increased from 30 to 100%, seedling growth of *Brassica napus* L. seeds decreased. Woodstock and Justice (1967) observed that a 0.5-kGy treatment of seeds stimulated the growth of the radish seedling, and 0.8 kGy inhibited growth. Chachin et al. (1972) observed that inhibitory effects of gamma radiation on the germination of radish seeds were reduced by treatment with high concentrations of gibberellic acid. Though there were many such studies, the control of either plant or human pathogens by irradiation was not studied until relatively recently. Cuero et al. (1986) investigated the influence of gamma irradiation and sodium hypochlorite sterilization on maize seed microflora and germination. The germination of the maize seed was not adversely affected by radiation doses up to 1.2 kGy, and the microflora were eliminated. Ramakrishna et al. (1991) compared sodium hypochlorite (12.5, 25, or 50%), mercuric chloride (0.1 or 0.2%), methyl bromide, propylene oxide, and gamma irradiation for their effectiveness in killing microorganisms on or within barley seeds. Gamma irradiation at 4 kGy eliminated most *Alternaria*, *Fusarium*, and *Epicoccum* spp., but 12 kGy was required to kill *Bacillus* spp. Germination was improved up to 8 kGy, but gradually decreased at doses up to 15 kGy.

• **Vegetables and Grains.** Cauliflower florets were packed in 15% CO_2 and irradiated at 2 kGy by Voisine et al. (1993). The process accelerated the deterioration of microsomal membranes during storage. Chervin and Boisseau (1994) replaced chlorination rinsing and spin-drying treatments of shredded carrots with irradiation at 2 kGy. Growth of aerobic and lactic microflora on the shredded carrots was inhibited by irradiation, and sensory analysis panelists preferred the irradiated vegetables. Sitton et al. (1995) irradiated wheat seed infested with teliospores of *Tilletia controversa* and *Tilletia tritici* with high-energy electrons (10 Mev). Doses of 4.6–6.7 kGy prevented germination of *T. tritici*; 10.2 kGy was required to eliminate germination of *T. controversa*. A dose of 1.2 kGy resulted in abnormal germination of the wheat.

Hayashi et al. (1997) examined the microbial load of rough rice, brown rice, wheat, and unhulled buckwheat following treatments with low-energy electrons from a Van de Graaff electron accelerator (180, 200, 225, 250, 300, and 500 kV and 8, 14, 22, and 40 A for 180 kV, 200 kV, 225 kV, and the other voltages, respectively). The seeds were irradiated on a vibrating table. The authors estimated that the average electron dose required to lower the microbial load of the seed to <100 cfu/g were 7–13, 15–20, and 7–12 kGy, for brown rice, rough rice, and wheat, respectively. The authors observed significant reductions in starch viscosity by a radiation dose of 10 kGy from gamma but not electrons, attributing the difference to the very low penetration of the seed by these low-energy electrons.

The use of low-energy electrons for food treatment was proposed by Nablo (1984). Hayashi (1998) and Hayashi et al. (1998) discovered that low-energy electrons (< 300 kEv) could decontaminate grains and dehydrated vegetables without penetration and degradation of starch.

The penetration of an electron beam depends on the accelerating voltage and density of the target material. Richards (1968) defined the maximum depth of penetration of an electron by the following equation:

$$Rm = (0.542 \times 10^{-6} V - 0.133) \div r$$

where V = accelerating potential and r = specific gravity of the target material.

Hayashi (1998) measured the penetration capacity of electrons at 60, 75, and 90 keV to be 6, 10, and 17.8 mg/cm^2, respectively. The very low penetration of the low-energy electrons accounts for the decreased effect on starch, but also limits their effectiveness when greater penetration is required.

Farkas et al. (1997) inoculated precut bell peppers and carrots with *L. monocytogenes*. The product was irradiated at 1 kGy and stored at 1–16°C. The irradiation treatment reduced the viable counts and those of *L. monocytogenes*. Hagenmaier and Baker (1997) reduced the normal microflora on commercially prepared fresh-cut lettuce by treating it with a radiation dose of 0.19 kGy. Eight days after irradiation, the unirradiated lettuce had 220,000 cfu/g, while the irradiated lettuce had 290 cfu/g. Hagenmaier and Baker (1998) compared irradiation (0.5 kGy) in an atmosphere of CO_2 to chlorination of shredded carrots. Nine days after treatment, the irradiated carrots had a microbial population of 1,300 cfu/g, compared to 87,000 cfu/g on the nonirradiated chlorinated controls.

A Promising Technology

Foodborne pathogens have been linked to several fruits, vegetables, and juices. In many cases, it is difficult if not impossible to either wash these pathogens off the produce or inactivate them by chemical treatments. This may be because the pathogens are located in inaccessible areas or they have penetrated the product.

Ionizing radiation can penetrate the entire product to inactivate pathogens that have penetrated the food. Pasteurization of juice is typically carried out by thermal processing with possible changes in flavor. Treatment with ionizing radiation may offer an alternative technique for some juice processors without changes in flavor. Treatment with ionizing radiation is a promising technology that can be used to improve the safety of ready-to-eat fruits and vegetables.

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